A NOTE ON AN UNUSUAL SPORE FORM IN *PUCCINIA MALVACEARUM* BERT.

By W. L. Waterhouse, The University of Sydney.

(Plate x, fig. 2.)

[Read 24th September, 1952.]

Puccinia malvacearum Bert., the cause of rust on hollyhocks and numerous plants in the family Malvaceae, is world-wide in its distribution.

Much work has been done on the rust in Europe and U.S.A. It is described as producing teleutospores only in its life history. Arthur (1934)* states, "Pycina unknown, probably not formed."

In class work the rust has been constantly used, both for studies of living and fixed material. In the latter, stained microtome sections of a rusted leaf of *Malva rotundifolia* L. which had been collected in the University grounds in 1937 showed the formation of a spermogonium on the upper surface of a teleutosorus (Plate x, fig. 2). At once a search was made for other occurrences, but without success. Much material has been examined in more recent times, but teleutosori only have been found.

In the case in point, which was shown to the late Professor F. T. Brooks, F.R.S., during his visit in 1939, the same mycelium can be seen to be producing the two spore forms. The fact that it was fixed made it impossible to carry out any cultural studies to determine the significance of the spermogonial formation.

Plate x, fig. 2.

Section through leaf of *Malva rotundifolia* showing formation of spermogonium above a teleutosorus. × 200.

* Arthur, J. C., 1934.—Manual of the rusts in the United States and Canada. Purdue Research Foundation, Lafayette, Ind., 438 pp.

A NOTE ON THE OCCURRENCE OF AN UNDESCRIBED RUST ON *CRYPTOSTEMMA CALENDULACEUM* (L.) R. BR.

By W. L. Waterhouse, The University of Sydney.

(Plate x, figs. 3, 4.)

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On each of three occasions, viz., in July, 1937, April, 1939, and August, 1941, a plant of *Cryptostemma calendulaceum* (L.) R. Br. was found in waste land in the Sydney metropolitan area showing rust on the leaves. In each case, careful search revealed that they were the only individuals showing infection amongst some hundreds of plants. An examination of various other species of plants in the neighbourhood failed to reveal the presence of any other rust in any stage of development. In other years no rusted plants have been found although searches have been made.

The first two collections showed only aecidia, but in the third, spermogonia were also present, intermingled with aecidia.

Detailed examinations gave the following results:

Spermogonia epiphyllous, usually occurring singly but sometimes in swollen areas, pale yellow, 1 to 3 mm. in diameter. Spermogonia subspherical, about 100μ in diameter (Plate x, fig. 3).

Aecidia amphigenous but mainly epiphyllous, solitary or grouped in swollen yellowish areas up to 6 mm. in diameter. Peridium pale with a recurved margin. Peridial cells, somewhat rhomboidal in section, 12 to 20μ across (Plate x, fig. 4). Aecidospores globoid or ellipsoidal with a thin colourless wall about 1μ thick and finely verrucose. Measurements of 100 spores taken at random gave a mean length of $16{\cdot}8 \pm 0{\cdot}2\mu$ with a range of 11–21μ, and a breadth of $14{\cdot}4 \pm 0{\cdot}1\mu$ with a range of 10–17μ.

Viable aecidiospores were used to inoculate leaves of the infected plants, which had been transplanted to pots, as well as a series of seedlings taken at random, but in no instance were infections obtained. Cineraria seedling inoculations also gave negative results.

Inquiries made at the Commonwealth Mycological Institute revealed that no rust on *Cryptostemma calendulaceum* had been recorded there. Similar inquiries sent direct to South Africa, where the plant is indigenous, brought the same reply.

The name *Aecidium cryptostemmatis* is proposed for the rust. Further investigations may reveal its full life history.

Plate x, figs. 3, 4.

Sections of leaf of *Cryptostemma calendulaceum* showing a typical spermogonium and aecidium. × 200.

STUDY OF SOIL ALGAE.

1. Fluorescence Microscopy for the Study of Soil Algae.

By Y. T. Tchan,

Macleay Bacteriologist to the Society.

[Read 24th September, 1952.]

Synopsis.

A new fluorescence microscopy technique for the estimation of soil algal population is proposed. A critical examination of the technique shows the advantages of the method over the classical dilution culture technique, and the limits of its possibilites are discussed.

Although the existence of an abundant soil algae flora has been recognized for many years, very few quantitative data on soil algae populations are available, even for soils in which the algal flora is known to be of great economic importance. This paucity of data is ascribed to the lack of an accurate and rapid technique for measuring algal populations.

In the account that follows, a new and rapid technique for the quantitative estimation of algae in the soil, based on the fluorescence of chlorophyll is described. The writer considers that the technique outlined below will provide a valuable tool for the study of algal floras, not only from the academic standpoint, but also from the economic, e.g., in rice fields and irrigated areas[1][4][6][8][17] which are becoming an increasingly important section in agriculture.

The application of fluorescence microscopy to biology is not new. J. Levaditi[10] gave a good idea of its importance in his review and in 1948 Struger[20] described the Orange-acridin to render bacteria visible under fluorescence microscopy. His technique, however, cannot be applied to soil algae because soil particles absorb the dye and can be confused with algae.

The technique described below uses the natural fluorescence of the chlorophylls to detect the soil algae. No dye is needed. The fluorescent algal cells are readily visible as red or orange-red images, according to the presence of associated pigments, against the dark background.

Equipment.

Light Source: An intense light source is required, and for critical work a special fluorescence lamp or arc lamp is necessary. For ordinary work, however, a low voltage lamp (6–12 volts) slightly overrun is satisfactory. A high aperture condenser lens is used to form a parallel beam. The frosted type of condenser must be rejected. The light is passed through a liquid filter made with $CuSO_4$ in aqueous ammonia according to the formula given by Augier.[2] To prevent the blue light entering the observer's eye, a yellow filter is used. (Ilford delta, Wratten, Zeiss stop filter, or Augier's liquid filter[3] are very satisfactory.)

Ordinary microscopes can be used if the condenser has an aperture of at least N.A.–1·2. Aperture of N.A.–1·4 is more desirable. An objective lens of 10 times magnification is used for routine work. Higher power lenses, including immersion lenses, can be used. The eyepiece should be of low magnification. For most routine work a 5× eyepiece is satisfactory. The high absorption of light in the prism system makes the binocular microscope unsuitable.

The Counting Chamber used is of hemacytometer type and can be made easily by sticking 2 cover slips (No. 0) with balsam on the surface of an ordinary microscope slide